Biochemical study of the effect of typhoid fever and brucellosis infections on some serum salts concentration

Al–Hadithi H. Amal*

Received 20/4/2004 ; accepted 16/8/2004

الخلاصة

تم دراسة عشرون حالة من مرضى حمى التيفوئيد والحمى الراجعة (حمى مالطا) سريرياً ومصلياً وبايوكيمياوياً . أهم ما وجد سريرياً إن معظم الحالات صاحبها حمى مستمرة (٦٦%) وكلما تقدم المرض كلما زادت أعراض الجهاز الهضمي وكمضاعفات صاحب الحالات نزف معوي (١٩%) . أما من الناحية المصلية فان عدد المرضى الذين ثبت وجود التيفوئيد لديهم بواسطة (Widal (test) كانوا ثلاثة عشر مريضاً والذين ثبت وجود حمى مالطا لديهم بواسطة (Rose Bengal test) خمسة مرضى والذين أعطوا نتيجة موجبة مع (Widal test) و (Widal test) الثان فقط ، وكذلك أجريت الدراسة على عشرة أشخاص أصحاء ظاهرياً نتيجة موجبة مع (Widal test) و (Widal test) الثان فقط ، وكذلك أجريت الدراسة على عشرة أشخاص أصحاء ظاهرياً كمجموعة للمقارنة . وجد من هذه الدراسة ان المعدل الكلي لمستوى مصل المغنيسيوم والكالسيوم الكلي مرتفع ظاهرياً عند مرضى الحمى التيفوئيدية (2001 و 0.00 P ع) وانخفاض ظاهري في مستوى مصل البوتاسيوم (2000 P ع) . . بينما مرضى حمى مالطا أظهروا ارتفاع قليل في المعدل الكلي لمستوى المعنوي مصل البوتاسيوم (2000 P ع) . . بينما مرضى حمى المعدل الكلي لمستوى مصل البوتاسيوم (2000 P ع) . أما مرضى حمى مالطا والتيفوئيد معاً أثبتوا ارتفاع ظاهري في المعدل الكلي معدل المعدل الكلي لمستوى مصل البوتاسيوم (2000 P ع) . أما مرضى حمى مالطا والتيفوئيد معاً أثبتوا ارتفاع ظاهري في المعدل الكلي المعدل الكلي لمستوى مصل البوتاسيوم (2000 P ع) . أما مرضى حمى مالطا والتيفوئيد معاً أثبتوا ارتفاع ظاهري في المعدل الكلي المعدل الكلي لمستوى مصل البوتاسيوم والكالسيوم الايوني و 0.000 P ع و 0.00 P عال المحيون التغيرات المعدل الكلي لمستوى مصل البوتاسيوم والكالسيوم الايوني في ممالطا والتيفوئيد معاً أثبتوا ارتفاع ظاهري في المعدل الكلي المعدل الكلي لمستوى مصل البوتاسيوم والكالسيوم الايوني الايوني ال حمى مالطا والتيفوئيد معاً أثبتوا الموي في المعدل الكلي لمستوى مصل المغنيسيوم ، الصوديوم والكالسيوم الايوني (0.00 P ع و 0.000 P ع و 0.000 P ع المور الحار التغيرات البيدف :

(أ) لدراسة العلاقة الأكثر احتمالية بين حمى مالطا والحمى التيفوئيدية بواسطة التحليل البايوكيمياوي لتركيز بعض الاملاح في مصول هؤلاء المرضى .

(٢) لأن الـ (Widal test) أصبحت أهميته محدودة في تشخيص مرضى الحمى التيفوئيدية لذلك استخدمت مصول هؤلاء المرضى لدراسة تركيز بعض الأملاح والنظر إذا كان بالامكان استخدامها كمرجع للتفريق بين المرضين .

ABSTRACT

Twenty cases of typhoid and brucellosis patients were studied on clinical, serological and biochemical aspects. The mean clinical findings were, most of the cases had sustained fever 66%, gastroenterial symptoms developed as the disease progressed and intestinal hemorrhage 19% as a complications. In serological findings, the number of patients who had +ve positive widal test were thirteen and those who had positive Rose Bengal test were five and those who had positive Widal and Rose Bengal test were two patients. The study was also conducted on ten apparently healthy individual as a control group.

The objective :

(1) To study the possible correlation between brucella infection and salmonella typhi infection by biochemical test of some serum salts, and

(2) Because of the limited value of Widal's test in the diagnosis of typhoid fever, serum salts of these patients studied to see if can be used as a recommended test to differentiate between these two diseases.

^{*} Department of Clinical Biochemistry, College of Pharmacy, Baghdad University, Baghdad - Iraq.

The biochemical findings were reveled a significant increase in the level of serum magnesium (P \leq 0.004) and a significant decrease in the level of serum potassium (P \leq 0.003) in typhoid fever patients. While patients with brucellosis show moderate increase in the mean value of serum magnesium and sodium (P \leq 0.1 and P \leq 0.08 respectively) and moderate decrease in the mean value of serum potassium (P \leq 0.08).While patients with typhoid and brucellosis (combination) show significant increase in the mean value of serum magnesium and sodium (P \leq 0.001 and P \leq 0.001 respectively) while there is increased in the level of serum total calcium (P \leq 0.1) in patient with typhoid fever only. While serum ionized calcium show significant increase in the mean value in patients with typhoid fever and brucellosis (P \leq 0.06). The changes in these biochemical parameters were well discussed .

INTRODUCTION:

Typhoid fever, is a severe systemic illness transmitted through food or water, which is caused by the bacterium salmonella typhi serotype AB, BO, and DO, which isolated from stool, blood or urine cultures⁽¹⁾. Salmonella species cause enterocolitis, enteric fevers such as typhoid fever, and septicemia with metastatic abscesses⁽²⁾. They are gram negative rods, their antigens-cell wall O, flagellar H, and capsular Vi (virulence) are important for taxonomic and epidemiologic purposes. The O antigens, which are the outer poly-secharide of the cell wall, are used to subdivide the salmonella in to group A1 and flagellar H and antigens are subdivided in to 2 phases, phase 1 and phase 2. The Vi antigens are used for the typing of salmonella typhi, the agent of typhoid fever⁽³⁾. The genus of salmonella are divided into three species : salmonella typhi, salmonella choleraesuis, and salmonella enteritidis . The three types of salmonella infections enterocolitis, enteric fevers, and septicemia have different pathogenetic features . Entero colitis: is characterized by an invasion of the epithelial and subepithelial tissue of the small and large intestines with resulting inflammation and diarrhea. In typhoid and other enteric fevers, infection begins in the small intestine but few gastrointestinal symptoms occur. The organism enter and spread to the phagocytic of the liver, gall bladder, and spleen, this lead to bacteremia. Septicemia accounts for only about 5-10% of salmonella infections⁽⁴⁾. Zhang in $(1997)^{(5)}$ used three metallic salts ferrous, magnesium and calcium to induce and prevent Vi-antigen of salmonella typhi loss, and showed that induction and prevention of Vi-antigen of salmonella typhi loss by the three metallic salts was higher than that of other reports.

Brucella organisms are facultative intracellular bacteria that may infect many species of animals as well as humans⁽⁶⁾. Brucella species cause brucellosis are small gram–negative rods without capsules . The three major human pathogens and their animal reservoirs are brucella melitensis (goats and sheep), brucella abortus (cattle) and brucella suis (pigs) . The organisms enter the body either by ingestion of contaminated milk products or through the skin by direct contact in an occupational setting such as an abattoir . They are localize in the reticuloendothelial system, namely the lymph nods, liver, spleen, and bonemarraw⁽⁷⁾. Malik in (1998)⁽⁸⁾ in his study showed that there is a high serum calcium in human brucellosis .

MTERIALS AND METHODS :

Blood samples were collected from the patients on admission, and their sera were separated and analyzed at the time of separation. To compare the results a normal control group of ten apparently healthy persons were selected. Blood samples were also collected, separated and analyzed at the time of separation. Biochemical parameters studied were serum levels of magnesium, sodium, potassium and calcium (total and ionized) and evaluated in twenty patients with typhoid fever, brucellosis and typhoid–brucellosis infection. Measurement of Serum Magnesium :

By magnesium calmagite kite in which Mg. reacts with calmagite to form a coloured compound at pH 12. The intensity of the colour is proportional to the magnesium concentration⁽⁹⁾.

Measurement of Serum Sodium and Potassium :

Measurement of serum sodium and potassium by automated flam photometer using (Technicon Microlyzer). In this method sodium (Na) and potassium (K) flam photometer Tr is used to simultaneously determine Na, and K concentration from the same sample⁽¹⁰⁾.

Measurement of serum calcium (total and ionized) :

By automated methylthymol blue method (technicon microlyzer). The method is based on the formation of a blue complex between calcium and methylthymol blue, sodium sulphite was used as a reducing agent to minimise fading of the color ⁽¹¹⁾.

Statistical Analysis :

Statistical analysis of the data obtained in this study were performed using students "t" test⁽¹²⁾ to examine the difference in the parameters between typhoid fever, brucellosis, and typhoid-brucellosis combination patients and control group.

RESULTS AND DISCUSSION :

Serum Magnesium :

The mean value of serum magnesium show a significant increase in patients with typhoid fever, and combination (typhoid–brucellosis) than the control group (P<0.004 + P<0.01 respectively) Table (1).

Chemical	No. of	S			
categories	subject	Mean mg/dL	S.D	S.E±	t.test
Control	10	1.92	0.179	0.05	-
Typhoid	13	2.16	0.24	0.06	$P \le 0.004$
Brucellosis	5	2.12	0.3	0.13	$P \le 0.1$
Typhoid & Brucellosis	2	2.38	0.1	0.07	$P \leq 0.001$

Table 1 . Determination of serum magnesium in typhoid fever, brucellosis and typhoid-brucellosis patients and control.

S.D. : Standard deviation

S.E. : Standard error

This results showed that the increase of serum magnesium lead to induction and prevention of Vi antigen of Salmonella typhi loss. This results were agree with the Zhang report (1997) who used three metallic salts (ferrous, magnesium and calcium) in the induction and prevention of Vi antigen of Salmonella typhi loss⁽⁵⁾. While in brucellosis there is a little effect on magnesium concentration by brucella.

Serum Sodium and Potassium :

The results of serum sodium showed a significant increase in the mean value in patients with brucellosis and combination (P<0.08, P<0.001 respectively) Table (2), While there is no change in patients with typhoid fever.

Chemical categories	No. of				
	subject	Mean mg/dL	S.D	S.E±	t.test
Control	10	134.7	6.3	2	-
Typhoid	13	136	12.6	3.5	N.S.
Brucellosis	5	140	6.9	3	$P \le 0.08$
Typhoid & Brucellosis	2	143.5	0.7	0.5	$P \le 0.00$

Table 2. Determination of serum Sodium in typhoid fever, brucellosis and typhoid-brucellosis patients and control.

S.D. : Standard deviation

S.E. : Standard error

The hypernatremia in those patients may be due to high fever, which lead to increase the concentration of serum sodium⁽¹³⁾. On the other hand, the results of serum potassium showed a significant decrease in the mean value in patients with typhoid fever and brucellosis than the control group (P<0.003 and P<0.08 respectively) Table (3). The hypokalemia in those patients may be due to excessive sweating, which lead to decrease serum potassium⁽¹⁴⁾.

Table 3	. Determination	of serum	Potassium	in typhoid	fever,	brucellosis a	and
typhoid-	brucellosis patie	ents and c	ontrol.				

Chemical	No. of	Se			
categories	subject	Mean mg/dL	S.D	S.E±	t.test
Control	10	4.53	0.33	0.1	-
Typhoid	13	4	0.53	0.5	$P \le 0.003$
Brucellosis	5	4	0.42	0.1	$P \le 0.08$
Typhoid & Brucellosis	2	4.3	0.7	0.5	N.S.

S.D. : Standard deviation

S.E. : Standard error

Serum Calcium (Total and Ionized) :

The serum total calcium showed no significant change in patients with brucellosis and combination, while there is moderate increase in the mean value of serum total calcium in patients with typhoid fever. (P<0.1) Table (4).

Table 4 . Determination of serum total Calcium in typhoid fever, brucellosis and typhoid-brucellosis patients and control.

Chemical	No. of	Se			
categories	subject	Mean mg/dL	S.D	S.E±	t.test
Control	10	9.3	0.61	0.17	-
Typhoid	13	9.6	0.64	0.2	$P \le 0.1$
Brucellosis	5	9.6	0.46	0.2	N.S.
Typhoid & Brucellosis	2	9.1	1	0.7	N.S.

S.D. : Standard deviation

S.E. : Standard error

While serum ionized calcium show a significant increase in the mean value in patients with typhoid fever and brucellosis (P<0.06 and P<0.06 respectively), and no significant change in the mean value of serum ionized calcium in patients with combination Table (5).

Chemical	No. of	Serum Ionized calcium mg/100ml				
categories	subject	Mean mg/dL	S.D	S.E±	t.test	
Control	10	0.57	0.19	0.05	-	
Typhoid	13	0.66	0.07	0.02	$P \le 0.06$	
Brucellosis	5	0.68	0.15	0.06	$P \le 0.06$	
Typhoid & Brucellosis	2	0.62	0.09	0.06	N.S.	

Table 5 . Determination of serum lonized calcium in typhoid fever, brucellosis and typhoid-brucellosis patients and control.

S.D. : Standard deviation

S.E. : Standard error

These results were agreed with Malik (1998). Who reported that there is a high serum calcium in human brucellosis. This hypercalcemia in brucellosis may be due to the intact of vertebra and bone by brucellosis species leading to bone resorption and increase the concentration of serum calcium⁽⁸⁾. While Zhang (1997)⁽⁵⁾ used calcium as metallic compound to induce and prevent the Vi antigen of salmonella typhi loss, so according to this result the increase serum ionized calcium level in patients with typhoid fever is required to mentain the growth of bacteria.

From all results presented in this study it appears that many of these parameters measured might be of value in the predicted and follow up of patients with typhoid fever and brucellosis, but can not be used as a recommended method in diagnosis due to the less number of patients in this study. Therefore a careful study of these parameters (salts) in two types of diseases is needed to underline the exact effect of these two diseases on these salts.

REFERENCES:

1. MEDLINE. Epidemic Typhoid Fever–Dushnabe, Tajikistan. MMWR –Morb–Mortal–Wkly–Rep. 1998. Sep. 18; 47 (36): 752–6.

2. Nguyen–B.M.; Lanata–CF; Black–RE; Gil–AL; Karncil–A; and Wretlind B. Age–related prevalence of Shigella and Salmonella antibodies and their association with diarrhoeal diseases in peruvian children. Scand–J–infect–Dis. 1998; 30 (2): 159–64. Sweden.

3. Vollum–RL; Jamison–D.G; and Gommins–C.S.; Fairbrothers coliform bacteria textbook of bacteriology. Tenth Ed. 1970; P. 208–220.

4. Warren–E. Levinson Ernest Jawetz Gram –negative rods related to the enteric tract medical microbiology and immunology examination and board review. International Ed. 1989; P. 80–95.

5. Zhang–JX; Cao–J.; Tu–J. The induction and the prevention of Vi antigen of S. Typhi loss by three metalic ions of salts, Chin, 1997; Apr; 18(2): 89–91.

6. Godfroid–F; Taminiau–B; Danese–I; and 6 others; Identification of the persamine synthetase gene of brucella melitensis 16M and involvement of lipopoly saccharide 0 side chain in brucella survival in mice and in macrophages. Infect. Immun. 1998 Nov; 66(11): 5485–93.

7. Warren–E. Levinson Ernest Jawetz. Gram–negative rods related to animal sources medical microbiology and immunology examination and board review 1989. P. 100–103.

8. Malik–GM. High serum calcium in human brucellosis: a case –control study. Am–J–Trop–Med. H/g. 1998 Sep.; 59 (3): 397–8. Saudia Arabia.

9. GINDLER E.M.; HETH D.A.; Clin. Chem. 1971; 17. 662.

10. VARLEY'S H. GOWENLOCK A.H. and BELL M. Practical clinical biochemistry, Vol.1, 5th Ed. 1980.

11. Gindler E.M.; and King J.D. Amer. J. Clin. Path. 1972; 58; 376.

12. Robert C.D.; Rebecca G.K.; and Miller M.C. Tests of hypothesis on population meals introductory biostatistics for the health sciences 2nd Ed. New York. 1983; P. 97.

13. SMITH A.F.; BECKETT G.J.; WALKER S.W.; and RAE P.W.H.; Water, sodium and potassium balance. 6th Ed. Lecture notes on Clin. Bio. 2000; P. 24.

14. SMITH A.F.; BECKETT G.J.; WALKER S.W.; and RAE P.W.H.; Abnormalities of plasma potassium concentration. 6th Ed. Lecture notes on Clin. Bio. 2000; P. 28.